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Applied nutritional investigation

Single pyruvate intake induces blood alkalization and modification of resting metabolism in humans

Robert A. Olek Ph.D. [a,*], Marcin Luszczyk Ph.D. [b], Sylwester Kujach M.Sc. [b], Ewa Ziemann Ph.D. [b], Magdalena Pieszko Ph.D. [c], Ivo Pischel Ph.D. [d,e], Radoslaw Laskowski Ph.D. [b]

[a] Department of Biochemistry, University of Physical Education and Sport, Gdansk, Poland
[b] Department of Physiology, University of Physical Education and Sport, Gdansk, Poland
[c] Department of Clinical Nutrition, Medical University of Gdansk, Gdansk, Poland
[d] Centre for Pharmacognosy and Phytotherapy, UCL School of Pharmacy, University of London, London, United Kingdom
[e] Phytolab GmbH & Co. KG, Vestenbergsgreuth, Germany

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ABSTRACT

Objectives: Three separate studies were performed with the aim to 1) determine the effect of a single sodium pyruvate intake on the blood acid-base status in males and females; 2) compare the effect of sodium and calcium pyruvate salts and establish their role in the lipolysis rate; and 3) quantify the effect of single pyruvate intake on the resting energy metabolism.
Methods: In all, 48 individuals completed three separate studies. In all the studies, participants consumed a single dose of pyruvate 0.1 g/kg 60 min before commencing the measurements. The whole blood pH, bicarbonate concentration, base excess or plasma glycerol, free fatty acids, glucose concentrations, or resting energy expenditure and calculated respiratory exchange ratio were determined. The analysis of variance for repeated measurements was performed to examine the interaction between treatment and time.
Results: The single dose of sodium pyruvate induced blood alkalization, which was more marked in the male than in the female participants. Following the ingestion of sodium or calcium pyruvate, the blood acid-base parameters were higher than in the placebo trial. Furthermore, 3-h post-ingestion glycerol was lower in both pyruvate trials than in placebo. Resting energy expenditure did not differ between the trials; however, carbohydrate oxidation was increased after sodium pyruvate ingestion.
Conclusion: Pyruvate intake induced mild alkalization in a sex-dependent fashion. Moreover, it accelerated carbohydrate metabolism and delayed the rate of glycerol appearance in the blood, but had no effect on the resting energy expenditure. Furthermore, sodium salt seems to have had a greater effect on the blood buffering level than calcium salt.
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Introduction

Pyruvate is involved in various energetic reactions that take place in the cells. Manufacturers of dietary supplements are marketing pyruvate worldwide as a compound that influences fat metabolism and accelerates weight and body fat reduction [1]. Initial research indicated a positive effect of the pyruvate supplementation used in a combination with dihydroxyacetone (DHA) [2–4]. Because pyruvate alone has been found to have comparable or greater effects on the body composition and the metabolic indices than in combination with DHA [5], the studies on solely pyruvate supplementation have been performed [6–8]. The early studies reported a significant decrease in body weight and body fat in obese individuals given large doses of pyruvate (22–44 g/d) supplemented over 3 to 6 wk [7,8]. These dosages

Ivo Pischel was an employee of PhytoLab GmbH & Co. KG, Germany until August 2013.

* Corresponding author. Tel: +48 58 5547 214; fax: +48 58 5522 911.
E-mail address: Robol@awf.gda.pl (R. A. Olek).

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were obtained by a combination of both sodium pyruvate (NaP) and calcium pyruvate (CaP) [7,8]. In regard to the fact that the commercially available supplements provide 0.5 to 1.0 g of pyruvate per serving and the recommended daily uptake is only a few grams, later studies examined the effectiveness of lower pyruvate dosages [9,10]. One study [9] reported that administration of CaP 6 g/d for 6 wk combined with training three times weekly (45–60 min at 60% of predicted maximal heart rate) induced a modest but significant decrease in body weight and body fat in the overweight individuals in comparison to a placebo.

Despite the fact that several studies have indicated that pyruvate supplementation may affect body composition, the mechanisms of this action are not fully understood. It has been proposed that pyruvate may influence appetite, energy intake, energy metabolism, and the efficiency with which ingested foods are used [8]. One theory proposed that pyruvate activates and/or enhances a futile cycle, resulting in excessive energy expenditure [7]. It has also been suggested that pyruvate increases the proportion of energy derived from fat [5].

Increased fat utilization can be altered by the accelerated hydrolysis (lipolysis) of triacylglycerol (TG) reserves. An excessive lipolysis leads to the use of free fatty acids (FFA) as oxidative fuels. The rate of lipolysis is controlled by several factors [11,12]; one of the most well-known factors is pH [13]. It has been demonstrated that plasma FFA concentration decreases during acidosis, whereas alkalosis results in an increase in plasma FFA [14,15]. Recently, the buffering properties of NaP have been widely reviewed [16]. Therefore, the objectives of the conducted studies were to determine whether single NaP intake modifies blood acid-base status in a sex-dependent manner, NaP and CaP induce metabolic alkalosis and consequently rise in plasma FFA and glycerol concentrations in a similar manner, and single pyruvate intake affects the resting energy metabolism.

Methods

The entire experiment consisted of three subsequent stages. All parts of the investigation were approved by the Bioethics Committee at the Medical University of Gdansk and performed in accordance with ethical guidelines. The study was described to the participants before obtaining their written consent. The diets were not strictly standardized but the participants were instructed by the dietitian to maintain their nutritional habits throughout the study, to consume a light breakfast in the morning of the test day, and to refrain from caffeine and alcohol intake 6 and 24 h before each test, respectively.

Study 1

Twenty-two healthy individuals volunteered to participate in the study (age 23.8 ± 0.3 y; height 174.4 ± 1.8 cm; weight 72.3 ± 3.0 kg). On the day of the experiment, 11 women (age 23.8 ± 0.2 y; height 168.9 ± 1.8 cm; weight 61.6 ± 2.1 kg) and 11 men (age 23.7 ± 0.4 y; height 179.9 ± 1.5 cm; weight 82.9 ± 3.1 kg) reported to the laboratory at 09:00 h. After a 60-min rest period, blood samples were taken and NaP was ingested. For the following 180 min, participants remained in a sitting position, and the capillary blood samples were taken every 30 min for the acid-base status analysis.

Study 2

Fourteen healthy subjects (10 men, 4 women) participated in this study (age 23.6 ± 0.3 y; height 178.7 ± 1.5 cm; weight 74.6 ± 2.7 kg). The volunteers were randomly assigned to receive a single dose of NaP, CaP, or placebo in a double-blind, crossover design, with at least a 3-d washout period between the two conditions. On the day of the experiment, the participants reported to the laboratory at 09:00 h. After a 60-min rest period, blood samples were taken and the appropriate capsules were ingested. For the following 180 min, participants remained in a sitting position, and the blood samples were taken every 60 min for the acid-base status determination and for the later plasma glucose, FFA, and glycerol concentration measurements.

Study 3

Twelve healthy volunteers (10 men, 2 women) participated in study 3 (age 24.2 ± 0.2 y; height 180.2 ± 0.8 cm; weight 84.2 ± 4.0 kg). The volunteers received a single dose of NaP or placebo in a double-blind, crossover design. On the day of the experiment, the participants reported to the laboratory at 08:00 h. After a 60-min rest period, the experimental protocol was started. The control resting energy expenditure (REE) was determined during the following hour. At 10:00 h, participants consumed appropriate capsules and maintained resting for the following 180 min. For REE analysis, expired gas was collected through a facemask to measure the oxygen uptake (VO_2 L/min^{-1}) and the carbon dioxide exhalation (VCO_2 L/min^{-1}) breath by breath. A portable respiratory gas analysis system and a non-protein indirect calorimetric test were used during all the measurements (Cortex MetaMax, 3 B; CORTEX Biophysik GmbH, Germany). The Cortex MetaMax was calibrated against an ambient air and a standard calibration gas mixture provided by the manufacturer (15% O_2 and 4% CO_2), and the volume calibrated before every exercise test session. VO_2 and VCO_2 data were converted to REE using the equation:

$$\text{REE}\left(\text{J}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}\right) = (3.9 \times VO_2 + 1.1 \times VCO_2) \times 4184$$ [17].

REE was estimated from a mean of 20 min of continuous gas sampling during every hour of the protocol.

Values of the measured VO_2 and VCO_2 also were used to calculate the respiratory exchange ratio (RER) and thus the estimation of carbohydrate or fat utilization.

Pyruvate ingestion

The participants in the previous studies used approximately 0.07 to 0.08 g/kg of pyruvate [9,10]; therefore, the participants in our study consumed a single dose of 0.1 g/kg of NaP or CaP (which is ~0.08 g/kg). In the placebo trials, glucose was used at the dose of 0.05 g/kg. The compounds were encapsulated in identical gelatin capsules. NaP was purchased from Sigma-Aldrich Co., Germany, and CaP (pure and stable Calcium Pyruvate Monohydrate) was obtained from PhytoLab GmbH & Co., Germany.

Measurements

Blood-acid base status

The blood samples were taken into heparinized capillary tubes (Siemens Healthcare Diagnostics Inc, Tarrytown, NY, USA) and were immediately analyzed for the blood gases, pH, hemoglobin (Hb), and the electrolytes using a Rapidpoint 400/405 (Siemens Healthcare Diagnostics Inc, Tarrytown, NY, USA). Additionally, the bicarbonate (HCO_3^-) was calculated from PCO_2 and pH values according to the Henderson–Hasselbalch equation, and the base excess (BE) was calculated according to the following equation:

$$\text{BE} = (1 - 0.014 \times [\text{Hb}]) \times \left(\left[HCO_3^-\right] - 24.8 + (1.43 \times [\text{Hb}] + 7.7) \times (\text{pH} - 7.4)\right).$$

Plasma metabolites

The blood for the glucose, FFA, and glycerol analysis was centrifuged at 1000*g* for 10 min, and separated plasma samples were frozen at −70°C for later analysis. Plasma glucose, FFA, and glycerol were determined using standard kits (Randox Laboratories Ltd, Crumlin, UK). Assays were performed using a Super Aquarius CE9200 spectrophotometer (Cecil Instruments Ltd, Cambridge, UK).

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